

Figure 1

1 MQTCPLAEPG HVSQALGTLL FLAASLSAQN EGWDSPICTE GVVSVSWGEN
51 TVMSCNISNA FSHVNIKLRA HGQESAIFNE VAPGYFSRDG WQLQVQGGVA
101 QLVIKGARDS HAGLYMWHLV GHQRNNRQVT LEVSGAEPQS APDTGFWFVP
151 AVVTAVFILL VALYMFAWYR CRCSQQRREK KFFLLEPQMK VAALRAGAQQ
201 GLSRASAELW TPDSEPTPRP LALVFKPSPL GALELLSPQP LFFYAADP*

Figure 2

5'12 promoter (1-195) and cDNA (196-2180) sequence

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1  ATTCCTGCTT CTTTTAGCGT GAACGCGGGT GCGGTGCCTC CCGTGAAATA
51  ATAAATTCAC CGTCACGCTT GTTGTGAACG CGGGTGGTTC CCGAAACTTG
101 GAGGCTTCCC GTAAACCCAG CTCCTTCCTC ATCTGGGAGG TGGGTCCC GC
      ↓
151 GCGGGTCCGC CGCCTCCTCC CTGGCCCCTC CCTCTCGTGT CTTTCATTTT
201 CCTGGGGCTC CGGGGCGCGG AGAAGCTGCA TCCCAGAGGA GCGCGTCCAG
251 GAGCGGACCC GGGAGTGTTT CAAGAGCCAG TGACAAGGAC CAGGGGCCCCA
301 AGTCCCACCA GCCATGCAGA CCTGCCCCCT GGCATTCCCT GGCCACGTTT
351 CCCAGGCCCT TGGGACCCCT CTGTTTGTGG CTGCCTCCTT GAGTGCTCAG
401 AATGAAGGCT GGGACAGCCC CATCTGCACA GAGGGGGTAG TCTCTGTGTC
451 TTGGGGCGAG AACACCGTCA TGTCTGCAA CATCTCCAAC GCCTTCTCCC
501 ATGTCAACAT CAAGCTGCGT GCCCACGGC AGGAGAGCGC CATCTTCAAT
551 GAGGTGGCTC CAGGCTACTT CTCCCGGGAC GGTGCGCAGC TCCAGGTTCA
601 GGGAGGCGTG GCACAGCTGG TGATCAAAGG CGCCCGGGAC TCCCATGCTG
651 GGCTGTACAT GTGGCACCTC GTGGGACACC AGAGAAATAA CAGACAAAGTC
701 ACGCTGGAGG TTTCAGGTGC AGAACCCAG TCCGCCCTG ACACTGGGTT
751 CTGGCCTGTG CCAGCGGTGG TCACTGCTGT CTTCATCCTC TTGGTCGCTC
801 TGGTCATGTT CGCCTGGTAC AGGTGCCGCT GTTCCCAGCA ACGCCGGGAG
851 AAGAAGTTCT TCCTCCTAGA ACCCCAGATG AAGGTCGAG CCCTCAGAGC
901 GGGAGCCCAG CAGGGCCTGA GCAGAGCCTC CGTGAACTG TGGACCCAG
951 ACTCCGAGCC CACCCCAAGG CCGCTGGCAC TGTTGTTCAA ACCCTCACCA
1001 CTGGAGCCCC TGGAGCTGCT GTCCCCCCA ACCCTTGTTT CCATATGCCG
1051 CAGACCCATA GCCGCCTGCA AGGCAGAGAG GACACAGGAG AGCCAGCCCT
1101 GAGTGCCGAC CTTGGGTGGC GGGGCTGGG TCTCTCGTCC CACCCGAGG
1151 GCACAGACAC CGGCTTGCTT GGCAGGCTGG GCCTCTGTGT CACCCACTCC

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1201 TGGGTGCGTG CAGACCCCTTC CCCTCCACCC CCCAGGTCTT CCAAGCTCTG
 1251 CTTCCTCAGT TTCCAAAATG GAACCACCTC ACCTCCGCAG CACCCGACTT
 1301 ACCAGGACGC ATGCCCTCC CTCTGCCCTC ATCAAACCCA CAGACCCGGA
 1351 CTCCTTTTCT GCCACCCAG GCTGGTCCGG CCCAGGTGT GGGGTCCGCT
 1401 CTCTCCACTC CCAGGGCTCC GCGCCCAAGT GAGGGGGCCC CTGCCGGAGC
 1451 CTCAGACACA CTGGAGTTCA GGGCTGGGGG GGCCTTGCA CATACCTGTC
 1501 CCTTGGCTAT GAGCAGGCTT TGGGGGCCCT TCCGCGGCAG CCCCGGGGGC
 1551 CGAGGTAGG TCTGGGGGCT TAGAGGCTGG GATGGCTCCT GGGCCACCG
 1601 CCAGGGGGCA AGCGCAGGCC GGGCTGGGAG GCGGCGGCG CGGCTCGGGC
 1651 TGGGGGGTCA GGTGGACGCT GCCTCCGGG CTGGTCGCG ATCCCTCAGT
 1701 CCCTCGGCCA CCCGGGGGTC GCTCCCTCGT GCCCACCGA CCTGCCGAGC
 1751 CTCTTTGGAC CCAGATCTGT TCATGCTTT GTCTTCGTCA CTGCGGCGGG
 1801 GCCCTTTGAT GTCTTCATCT GTATGGGGT GAAAAATCAC CGGGAATCCC
 1851 CCTTCAGTTC TTTGAAAAAG TTCCATGACT CGAATATCTG AAATGAAGAA
 1901 AACAAACCGA CTCACAAACC TCCAAGTAGC TCCAAATGCA ATTTTAAAA
 1951 TGGAAAAACA AAATCTGAAA GAAACGTCTT TAGTGGCTTT AAGCCCCAAA
 2001 ACGTCCCTAA GCGCTCCTCG AGATGAAGAC GGGGGGGAGC CCCAGCCAGG
 2051 TGGAGACCCC GCAGGACGCG GCGGCGCCCG GTGACCGAGG CCTCGCACAG
 2101 CCGGCCGCCC TGAGGGTCGG GCCGAGCCAG GGTCCAAGAG GGGCGCGTTT
 2151 GTGTCTCGGG TTAAAATAAG GTTCCGTCCG

Figure 3 : K12 Expression

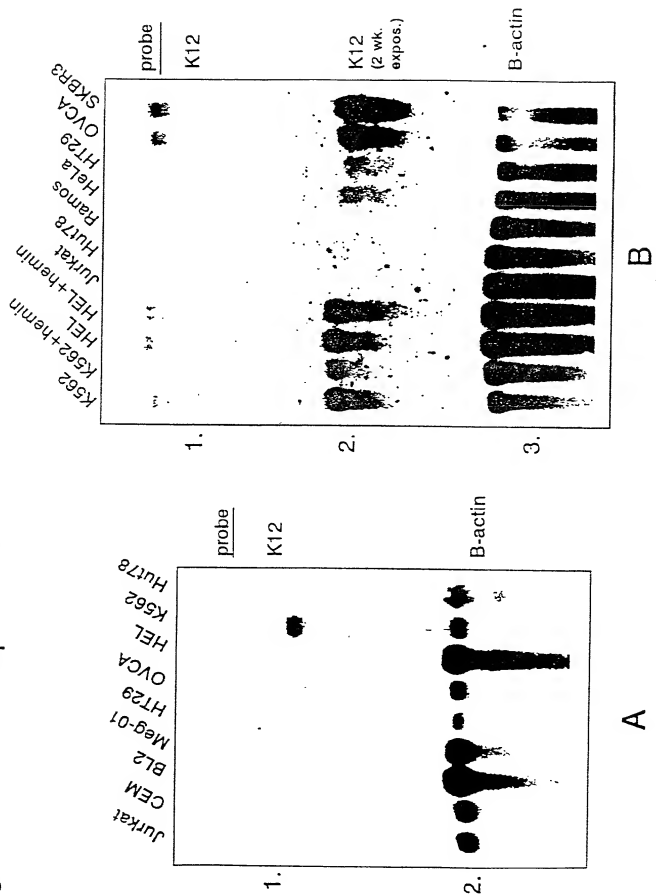
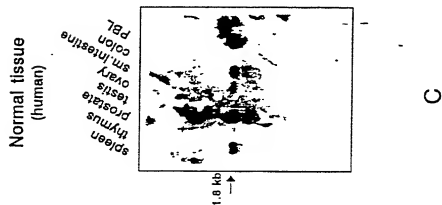
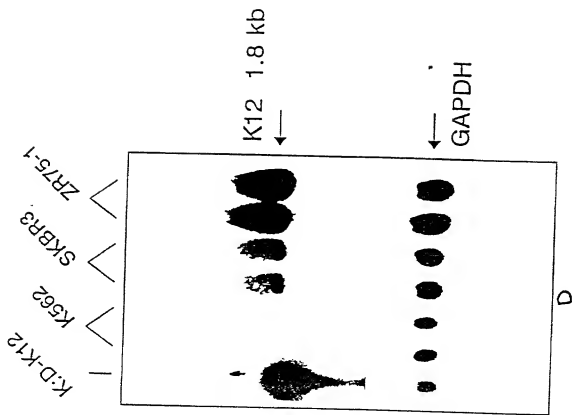


Figure 3 (cont)

Total RNA Northern



Western Blot Probed with Antiserum to K12

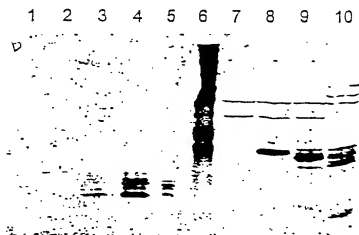


Figure 4. Western blot probed with antiserum to K12. Concentrated media from K562 cells transfected with:

- 1) empty vector
- 2) K12 and 7 amino acid flag
- 3) K12 with C terminus addition
- 4) Full length K12
- 5) ZR75-1 cells (not transfected)
- 6) Molecular weight standards (smallest is 32 kDa)

Soluble protein extracts from K562 cells transfected with :

- 7) empty vector
- 8) K12 with 7 amino acid flag
- 9) Full length K12
- 10) ZR75-1 cells (not transfected)

Figure 5: Subcellular Localization of K12 to the Golgi

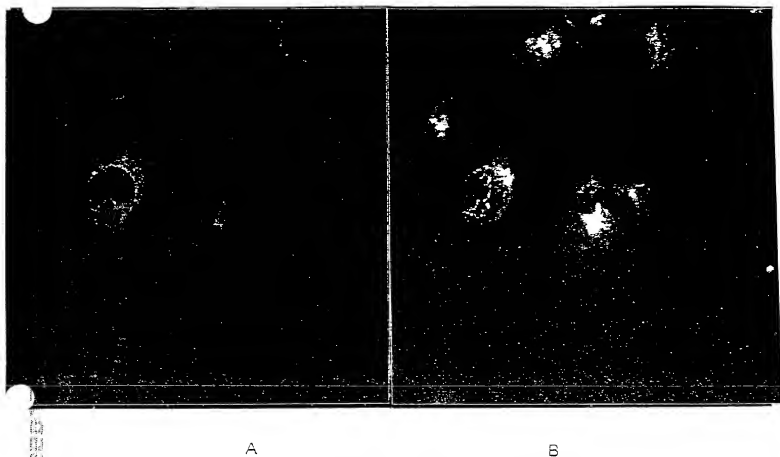


Figure 5

Same field of view of ZR75-1 cells that were grown on slides, acetone-fixed and double stained with,

A : antigen -purified anti-K12 polyclonal antibody followed by FITC-conjugated goat anti-rabbit IgG secondary antibody.

B: Rhodamine conjugated Wheat Germ Agglutinin (an immunochemical marker for Golgi bodies)

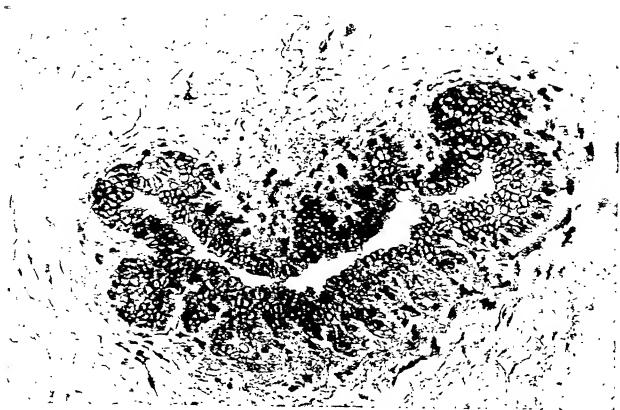


Figure 6 Immunoperoxidase staining of normal breast tissue, A, and colloid breast carcinoma, B, with monoclonal antibody 7C3 against K12. Panel C is a isotype matched P3 control. Dark brown staining reflects monoclonal antibody binding to K12 antigen.

Conditioned Media Proliferation Assay

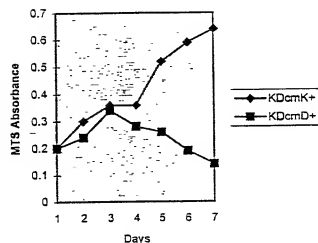


Figure 7: Growth Curves for K562 cells grown in conditioned media from :

KDcmK+, K562 cells secreting K12 into the media, or

KDcmD+, K562 cells transfected with an empty vector and producing no detectable K12 in media.